

We claim:

1. A method of producing an engineered blood vessel, comprising:
  - a. incorporating at least endothelial cells (ECs) and smooth muscle cells (SMCs) in a matrix;
  - 5 b. circumferentially positioning the matrix on the outer surface of a tubular support, wherein the support allows movement of mitoattractant, attractant, and mitogenic factors from within the support to said ECs; and
  - c. allowing movement of one or more mitogenic with one or more  
10 attractant factors or one or more mitoattractant factors or combinations thereof present within the support to said ECs, such that a bilayer is formed, said bilayer comprising a layer of said ECs and a layer of said SMCs;thereby producing an engineered blood vessel.
- 15 2. A method of making an engineered blood vessel comprising an endothelial intimal layer surrounded by a smooth muscle medial layer, said method comprising contacting one or more mitogenic with one or more attractant factors or one or more mitoattractant factors or combinations thereof with a matrix, said matrix  
20 having incorporated therein at least ECs and SMCs, said matrix and cells being circumferentially positioned around a tubular support, said factors having been added to the inside of the tubular support, said support having allowed said factors to move from the inside of the tube to the ECs in the matrix, wherein said contacting results in the formation of said endothelial intimal layer surrounded by said smooth  
25 muscle medial layer.
3. The method of claim 2, wherein the ECs are derived from stem cells.
4. The method of claim 3, wherein the stem cells are selected from the  
30 group consisting of embryonic stem cells, embryonic germ cells, multipotent adult progenitor cells (MAPCs), hematopoietic stem cells, mesenchymal stem cells, and endothelial progenitor cells.
5. The method of claim 2, wherein the SMCs are derived from stem  
cells.

6. The method of claim 5, wherein the stem cells are selected from the group consisting of embryonic stem cells, embryonic germ cells, MAPCs, mesenchymal stem cells, and smooth muscle progenitor cells.
7. The method of claim 3 or 5, wherein the stem cells are MAPCs.
- 5 8. The method of claim 3 or 5, wherein the stem cells are autologous.
9. The method of claim 3 or 5, wherein the stem cells are heterologous.
10. The method of claim 2, wherein the ECs are derived from vascular tissue.
- 10 11. The method of claim 2, wherein the SMCs are derived from vascular tissue.
12. The method of claim 10 or 11, wherein the vascular tissue is selected from the group consisting of pulmonary artery, pulmonary vein, femoral artery, femoral vein, saphenous artery, saphenous vein, iliac artery, iliac vein, umbilical artery, umbilical vein, microvascular tissue, and aortic tissue.
- 15 13. The method of claim 3 or 5, wherein the stem cells are derived from bone marrow, brain, spinal cord, umbilical cord blood, liver, muscle, fat or placenta.
14. The method of claim 2, wherein the matrix is comprised of a substance selected from the group consisting of fibrin, collagen, amphiphilic di-block copolymers, amphiphilic tri-block copolymers, and peptides.
- 20 15. The method of claim 2, wherein the matrix is comprised of fibrin.
16. The method of claim 2, wherein the support is comprised of porous plastic.
17. The method of claim 16, wherein the porous plastic is selected from the group consisting of polyethylene, a polyethylene derivative, polycarbonate, polylactic acid, and polyglycolic acid.
- 25 18. The method of claim 2, wherein the support further comprises agarose.
19. The method of claim 2, wherein the support comprises an internal matrix.
- 30 20. The method of claim 2, wherein the factor is vascular endothelial growth factor.
21. The method of claim 2, wherein the composition further comprises a basement membrane and a lumen.

22. The method of claim 1, further comprising:
- d. coating the bilayer with fibroblasts.
23. The method of claim 22, wherein the fibroblasts are derived from stem cells.
- 5 24. The method of claim 23, wherein the stem cells are selected from the group consisting of embryonic stem cells, embryonic germ cells, and MAPCs.
25. The method of claim 22, wherein the stem cells are MAPCs.
26. An engineered blood vessel produced by the method of claim 1.
27. An engineered blood vessel produced by the method of claim 2.
- 10 28. A composition in vitro comprising a matrix containing incorporated ECs and SMCs, said matrix containing said incorporated cells being circumferentially positioned around a tubular support, said support allowing movement of mitogenic, attractant, and mitoattractant factors across the support to said ECs, said composition comprising one or more mitogenic with one or more
- 15 attractant factors or one or more mitoattractant factors or combinations thereof within said support.
29. The composition of claim 28, wherein the ECs are derived from stem cells.
30. The composition of claim 29, wherein the stem cells are selected
- 20 from the group consisting of embryonic stem cells, embryonic germ cells, MAPCs, hematopoietic stem cells, mesenchymal stem cells, and endothelial progenitor cells.
31. The composition of claim 28, wherein the SMCs are derived from stem cells.
32. The composition of claim 31, wherein the stem cells are selected
- 25 from the group consisting of embryonic stem cells, embryonic germ cells, MAPCs, mesenchymal stem cells, and smooth muscle progenitor cells.
33. The composition of claim 29 or 31, wherein the stem cells are MAPCs.
34. The composition of claim 29 or 31, wherein the stem cells are
- 30 autologous.
35. The composition of claim 29 or 31, wherein the stem cells are heterologous.

36. The composition of claim 28, wherein the ECs are derived from vascular tissue.

37. The composition of claim 28, wherein the SMCs are derived from vascular tissue.

5 38. The composition of claim 36 or 37, wherein the vascular tissue is selected from the group consisting of pulmonary artery, pulmonary vein, femoral artery, femoral vein, saphenous artery, saphenous vein, iliac artery, iliac vein, umbilical artery, umbilical vein, microvascular tissue, and aortic tissue.

39. The composition of claim 29 or 31, wherein the stem cells are derived  
10 from bone marrow, brain, spinal cord, umbilical cord blood, liver, muscle, fat or placenta.

40. The composition of claim 28, wherein the matrix is comprised of a substance selected from the group consisting of fibrin, collagen, amphiphilic di-block copolymers, amphiphilic tri-block copolymers, and peptides.

15 41. The composition of claim 28, wherein the matrix is comprised of fibrin.

42. The composition of claim 28, wherein the support is comprised of porous plastic.

43. The composition of claim 42, wherein the porous plastic is selected  
20 from the group consisting of polyethylene, a polyethylene derivative, polycarbonate, polylactic acid, and polyglycolic acid.

44. The composition of claim 28, wherein the support further comprises agarose.

45. The composition of claim 28, wherein the support comprises an  
25 internal matrix.

46. The composition of claim 28, wherein the factor is vascular endothelial growth factor.

47. The composition of claim 28, wherein the composition further comprises a basement membrane and a lumen.

30 48. The composition of claim 28, wherein the composition further comprises a layer of fibroblasts.

49. The composition of claim 28, further comprising an outer casing surrounding the matrix.

50. A method of providing a vascular graft to a subject in need thereof, comprising providing the composition of claim 28 to the subject.
51. The method of claim 50, wherein the subject is a human.
52. The method of claim 50, wherein the vascular graft is vasoactive.
- 5 53. An engineered blood vessel comprising an intimal layer of ECs incorporated in a matrix and a medial layer of SMCs incorporated in a matrix, said layers being circumferentially positioned around a tubular support.
54. A pharmaceutical composition comprising an engineered blood vessel in a pharmaceutically acceptable carrier, said engineered vessel comprising an  
10 intimal layer of ECs incorporated in a matrix and a medial layer of SMCs incorporated in a matrix, said layers being circumferentially positioned around a tubular support.
55. An in vitro composition comprising ECs and SMCs incorporated in a matrix circumferentially positioned around a tubular support, wherein one or more  
15 mitogenic with one or more attractant factors or one or more mitoattractant factors or combinations thereof capable of permeating the support are present within the support.
56. The composition of claim 28 or 55, wherein the factor(s) is selected from the group consisting of FGF-1, FGF-2, FGF-4, angiogenin, angiopoietin,  
20 angiotensin, endothelin, acetyl-N-Ser-Asp-Lys-Pro, Angiomodulin, Angiotropin, endothelial cell growth factor, B61, endothelioma-derived motility factor, epidermal growth factor, endothelial cell-viability maintaining factor, IGF-1, heparin-binding neurotrophic factor, human uterine angiogenesis factor, platelet-derived endothelial cell growth factor, platelet-derived growth factor, Placenta growth factor/vascular  
25 permeability factor, transferrin, transforming growth factor-beta, interleukin-8, and growth hormone.
57. A method of culturing cells in a matrix comprising the steps of
- a. combining ECs and SMCs in a matrix;
  - b. growing ECs and SMCs in a matrix on the exterior surface of a  
30 tubular support, wherein the support allows movement of mitogenic, attractant, and mitoattractant factors from within the support to said ECs; and

- c. allowing movement of one or more mitogenic with one or more attractant factors or one or more mitottractant factors or combinations thereof present within the support to said ECs.

58. The method of claim 1, 2, or 57, wherein the factor(s) is selected  
5 from the group consisting of FGF-1, FGF-2, FGF-4, angiogenin, angiopoietin, angiotensin, endothelin, acetyl-N-Ser-Asp-Lys-Pro, Angiomodulin, Angiotropin, endothelial cell growth factor, B61, endothelioma-derived motility factor, epidermal growth factor, endothelial cell-viability maintaining factor, IGF-1, heparin-binding neurotrophic factor, human uterine angiogenesis factor, platelet-derived endothelial  
10 cell growth factor, platelet-derived growth factor, Placenta growth factor/vascular permeability factor, transferring, transforming growth factor-beta, interleukin-8, and growth hormone.